REPORT FOR NFWF RIVER HERRING WORKING GROUP

Taking stock of river herring: Population genetic structure in anadromous alewife and blueback herring

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Summary: We investigated population genetic structure in alewife and blueback herring using microsatellite markers deployed on samples collected from across the USA ranges of both species. Pairwise tests of genic differentiation show that natal homing behavior in both alewife and blueback herring has caused significant genetic differentiation among the majority of spawning runs sampled. Results from Bayesian clustering methods indicate the presence of higher-level genetic structure (i.e., stocks) within the alewife and blueback herring ranges. Specific sock boundaries are not congruent between the two species. We infer the presence of a minimum of 3 genetically distinguishable stocks in alewife: a **Northern New England Stock** [East Machias (ME), St George (ME), Lamprey (NH)], a **Southern New England Stock** [Mystic (MA), Town Brook (MA), Monument (MA), Gilbert Stuart (RI), Thames (CT), Bride Brook (CT), Connecticut (CT), Quinnipiac (CT), Housatonic (CT), Mianus (CT), Hudson (NY)], and a **Mid-Atlantic Stock** [Delaware (NJ), Nanticoke (MD), Rappahannock (VA), Chowan (NC), Roanoke (NC), Alligator (NC)]. We infer the presence of a minimum of 4 genetically distinguishable stocks in blueback herring: a **Northern New England Stock** [East Machias (ME), St George (ME), Exeter (NH)], a **Southern New England Stock** [Mystic (MA), Monument (MA), Gilbert Stuart (RI)], a **Mid-Atlantic Stock** [Connecticut (CT), Hudson (NY), Delaware (NJ), Nanticoke (MD), Rappahannock (VA), James (VA), Chowan (NC), Roanoke (NC), Neuse (NC)], and a **Southern Stock** [Cape Fear (NC), Santee-Cooper (SC), Savannah (GA), Altamaha (GA), St Johns (FL)]. We found a significant pattern of isolation by distance (IBD) in both species, although IBD was stronger in alewife compared to blueback herring. Thus, straying increases gene flow in blueback herring relative to alewife.

Introduction

River herring, the name used to collectively refer to alewife (*Alosa pseudoharengus*) and blueback herring (*Alosa aestivalis*), and are anadromous fishes that are native to the Atlantic Coast of North America. These species migrate from the ocean into coastal streams and rivers each spring to spawn. River herring are experiencing range-wide declines, which have become particularly dramatic over the past two decades (Limburg and Waldman 2009, ASMFC 2012). The goal of this project was to employ molecular genetic markers on alewife and blueback herring spawning runs throught the USA ranges of both species to explore (1) genetic divergence among spawning runs, (2) higher-level population structure indicative of genetically distinct spawning stocks at a larger geographic scale, and (3) the overall effect of geography on patterns of genetic divergence among spawning runs. Genetic markers have been used with great success to identify management units for a diversity fish species (reviewed in Waples et al. 2008). In particular, genetic methods for identifying population structure and designating management units have been integrated into the management of anadromous salmonids with great success (Waples 1995, Ford 2004) and have similar potential for anadromous alosines, including river herring (Hasselman and Limburg 2012).

Materials and Methods

Genetic markers and sampling: In order to investigate population genetic structure in alewife and blueback herring, we deployed 15 novel microsatellite markers on samples collected from across the USA ranges of both species. These genetic markers were developed specifically for alewife and blueback herring and were screened for variability throughout the ranges of both species before deployment (Labbe et al. 2012). We genotyped a total of 889 alewife samples from 20 rivers spanning the alewife range from Maine to North Carolina (Table 1). We genotyped a total of 1183 blueback herring samples from 20 rivers spanning the blueback herring range from Maine to Florida (Table 2).

Data conformance to model assumptions: Genotyping artefacts were assessed using MICROCHECKER v.2.2.3 (van Oosterhout *et al.* 2004). Tests for departures from Hardy-Weinberg equilibrium (HWE) and linkage disequilibrium (LD) were performed with GENEPOP v. 4.0.6 (Rousset 2007) using default parameters for all tests. Sequential Bonferroni adjustments were used to judge significance levels for all simultaneous tests (Holm 1979; Rice 1989). Selective neutrality of the microsatellite markers used in this study was evaluated using relative variance in repeat number (lnRV) and heterozygosity (lnRH) (Schlötterer 2002; Schlötterer and Dieringer 2005), and LOSITAN (Beaumont and Nichols 1996; Antao et al. 2008).

Genetic diversity: For each river the number of alleles per locus (Na), observed heterozygosity (H_O), an unbiased estimate of expected heterozygosity (H_E; Nei 1978), and F_{IS} (Weir and Cockerham 1984) were calculated using GENETIX v. 4.05 (Belkhir *et al.* 2004). Allelic richness (R) per locus was calculated for each river using FSTAT 2.9.3.2 (Goudet 1995; 2001) standardized to a minimum sample size of 24 individuals for alewife, and 26 individuals for blueback herring (LeBerg 2002).

Genetic differentiation: The statistical power and realized α-error for testing the null hypothesis of genetic homogeneity among rivers was assessed using POWSIM (Ryman and Palm 2006). Allelic heterogeneity among rivers was assessed via genic tests in GENEPOP v.4.0.6 (Rousset 2007) using default parameters for all tests. Tests were combined across loci or collections using Fisher's method. Hierarchical AMOVA was conducted to partition components of genetic variation among rivers, among collections, and among individuals within collections, using a permutation procedure (10,000 iterations) (Arlequin 3.1; Excoffier *et al.* 2005).

Overall and pairwise F_{ST} values (θ; Weir and Cockerham 1984) were estimated using FSTAT (Goudet 1995; 2001). The effect of variation in genetic diversity on genetic differentiation (Hedrick 2005) was accounted for by calculating standardized estimates of differentiation (F'_{ST}) using RECODEDATA v. 0.1 (Meirmans 2006) together with FSTAT to estimate $F_{ST(max)}$ for each pairwise comparison. Standardized estimates of differentiation were then calculated as $F'_{ST} = F_{ST} / F_{ST(max)}$ following Hedrick (2005).

Relationships among populations: Genetic affinities among rivers were examined using principal coordinates analysis (PCoA) of the pairwise genetic distance matrix for D_A (Nei *et al.* 1983) implemented in GenAlEx v.6.0 (Peakall and Smouse 2006).

Population structure: Two Bayesian model-based clustering methods, implemented in STRUCTURE v. 2.3.3 (Pritchard *et al.* 2000; Falush *et al.* 2003) and BAPS v. 5.3 (Corander *et al.* 2006), respectively, were used concomitantly in a hierarchical approach to infer the number of genetically homogenous clusters among rivers (Latch *et al.* 2006). For STRUCTURE, a burn-in of 50,000 replicates was followed by 250,000 replicates of the Markov chain Monte Carlo (MCMC) simulation, employing the admixture model and correlated allele frequencies among populations. Three iterations of this parameter set were performed for K (number of clusters) from 1-13, allowing an estimation of the most likely number of clusters. Both the plateau of likelihood values (Pritchard *et al.* 2000) and ΔK (*i.e.,* second order rate of change between successive K values; Evanno *et al.* 2005) were estimated.

For BAPS, the mixture model was first applied to cluster groups of individuals based on their multilocus genotypes. Three iterations of K (1-13) were conducted among populations to determine the number of genetically homogeneous groups. Admixture analysis was then conducted to estimate individual admixture proportions with regards to the most likely number of K clusters identified (Corander and Marttinen 2006), and visualized using DISTRUCT v. 1.1 (Rosenberg 2004).

Isolation by distance: Analysis of isolation by distance (IBD) was conducted among rivers to test for correlations between geographic distance and genetic differentiation using 10,000 permutations of the Mantel test implemented in IBDWS v. 3.15 (http://www.ibdws.sdsu.edu) (Jensen *et al.* 2005). Pairwise F'_{ST} values were linearized ($F'_{ST}/(1- F'_{ST})$) following Rousset (1997). Geographic distance between river mouths was measured using the Gebco 1-minute global bathymetry grid to identify land and ocean pixels. A Multistencil Fast Marching Method algorithm implemented in Matlab was then used to find the distances from each river mouth to each other pixel on the globe. The shortest path distance between river mouths was then calculated by summing the Euler distances for each pixel step and converting from degrees to kilometers.

Results

Data conformance to model assumptions: Evidence for null alleles resulted in the exclusion of loci for both alewife (*Aa082*, *Ap037, Ap047, Ap070*) and blueback herring (*Aa081, Ap058*) prior to further analyses (Microchecker). Remaining loci were retained as evidence for null alleles was sporadically distributed among loci and rivers. Exact tests revealed that genotypic frequencies were largely in accordance with HWE for both species ($p>0.05$; sequential Bonferroni correction for 20 comparisons). HWE departures for alewife and blueback herring remained for 11 and 20 locus-river comparisons, respectively, and were due to heterozygote deficiencies from sporadic null alleles. Exact tests of LD revealed that loci were physically unlinked and statistically independent ($p>0.05$; sequential Bonferroni correction for 1100 and 1560 comparisons for alewife and blueback herring, respectively). Relative variance in repeat number (lnRV) and heterozygosity (lnRH) failed to detect outlier loci for either species, and provided no evidence of non-neutrality.

Genetic diversity: Genetic polymorphism varied for both alewife and blueback herring depending on the locus and river considered. For alewife, the number of alleles per locus ranged from five (*Aa046*) to 19 (*Ap010*), with 8 loci exhibiting $\geq$9 alleles. H_O varied from 0.50 (Town Brook) to 0.67 (Delaware River), and *R* from 4.00 (Lamprey River) to 5.49 (Rappahannock River). Private alleles were observed for 13 populations at one or more loci, but were rare in frequency (<0.05; data not shown). For blueback herring, the number of alleles per locus ranged from seven (*Aa091, Ap047*) to 28 (*Ap037*), with 8 loci exhibiting $\geq$10 alleles. H_O varied from 0.50 (Gilbert-Stuart) to 0.57 (Nanticoke River), and *R* from 4.59 (Monument River) to 6.81 (Delaware River). Private alleles were observed for 16 populations at one or more loci, but were rare in frequency (<0.05; data not shown).

Genetic differentiation: An assessment of statistical power indicated that our microsatellite loci provided sufficient resolution to detect weak differentiation among alewife and blueback herring populations. The probability of obtaining a significant (p<0.05) result in contingency tests among populations with an F_{ST} of 0.001 was 0.86 and 0.98 (χ^2) for alewife and blueback herring, respectively, while maintaining the realized α-error at the intended level (0.05) for tests of genetic homogeneity.

Significant (p<0.05) genic differentiation between alewife populations was observed for 179/190 pairwise comparisons, with non-significant comparisons occurring among neighboring and geographically proximal populations (Table 3). Significant (p<0.05) genic differentiation between blueback herring populations was observed for 178/190 pairwise comparisons, with non-significant comparisons occurring predominately among rivers in the centre of the species' range (Table 4).

Standardized pairwise estimates of genetic differentiation (F'_{ST}) among alewife ranged from -0.003-0.352 (F_{ST}= -0.002-0.148); multilocus global F'_{ST}=0.119 (F_{ST}=0.049). Non-significant (p>0.05) genetic differentiation was observed among pairwise comparisons of neighboring and geographically proximal alewife populations. For blueback herring, F'_{ST} ranged from -0.008-0.233 (F_{ST}= -0.003-0.106); multilocus global F'_{ST}=0.067 (F_{ST}=0.030). Non-significant p>0.05) genetic differentiation was observed predominately (27/28) among pairwise comparisons of blueback herring populations in the center of the species' range.

For both species, hierarchical AMOVA revealed a significant (p<0.05) proportion of genetic variance partitioned among populations, and among individuals within populations. Non-significant variation among temporal replicates for both alewife and blueback herring suggested stable population structure over at least short (*i.e.*, 1-2 year) temporal scales.

Relationships among populations: PCoA revealed three factors that explained 92.25% of the variation in genetic distance (D_A) among alewife populations. Axis-1 explained 62.66% of this variation, and linear regression revealed a significant (r^2=0.85; p<0.001) relationship with latitude. Interestingly, alewife from the southern portion of their range (*i.e.,* Roanoke, Chowan, and Alligator Rivers) and from northern New England (*i.e.*, East Machias, St. George, and Lamprey Rivers) each clustered together, but away from populations near the centre of the species range. Three factors explained 85.66% of the variation in genetic distance (D_A) among blueback herring populations. Axis-1 explained 49.40% of this variation, and linear regression revealed a significant (r^2=0.81; p<0.001) relationship with latitude. The St. John's River population clustered well away from the remaining populations examined.

Population structure: For alewife, the maximum value of lnPr(X|*K*) using STRUCTURE was observed at *K*=4 (-24465.20). However, this estimate was only slightly greater than when K=3 (-24470.13), but had considerably more variation; suggesting that K=3 was more accurate. BAPS corroborated this result with significant (p<0.001) support for three genetically distinguishable clusters. Both methods identified the

same three clusters (*i.e.*, Northern New England, Southern New England, and Mid-Atlantic) (Figure 1), and further investigation using hierarchical STRUCTURE (Vaha *et al.* 2007) and BAPS analyses failed to detect additional structure within any of these clusters. Estimates of Δ*K* revealed the largest increase in the likelihood of the number of clusters at *K*=2, and suggested 'deep -rooted' structure among the 20 alewife populations surveyed. AMOVA revealed more variation among the three clusters (4.70%; p<0.001) than among rivers within clusters (1.30%; p<0.001). However, the detection of significant variation among rivers within clusters was consistent with the significant genic differentiation detected among most populations.

For blueback herring, the maximum value of lnPr(X|*K*) using STRUCTURE was observed at *K*=6 (-35108.260). However, this estimate was only slightly greater than when K=4 (-35189.77), or K=5 (-35163.20). BAPS had difficulty resolving population structure and provided nearly equivalent support for either K=4 (p=0.503) or K=5 (p=0.497). However, the greater variation in estimates for K=5 suggests four clusters across the U.S. range for blueback herring. Both STRUCTURE and BAPS identified the same four clusters (*i.e.*, Northern New England, Southern New England, Mid-Atlantic, and Southern) (Figure 2). Further investigation using hierarchical STRUCTURE and BAPS analyses failed to detect additional structure within any of these clusters. Estimates of Δ*K* revealed the largest increase in the likelihood of the number of clusters at *K*=2, and suggested 'deep -rooted' structure among the populations surveyed. AMOVA revealed substantially more variation among the four clusters (3.23%; p<0.001) than among rivers within clusters (0.22%; p<0.001), and was comparable to the among river component of variation (3.21%, p<0.05) when populations were not grouped into clusters. That AMOVA detected significant variation among rivers within clusters was consistent with the significant genic differentiation observed among most populations sampled.

Isolation by distance: Mantel tests revealed a highly significant (p<0.001) pattern of IBD for both alewife (r=0.73) and blueback herring (r=0.71) across their U.S. range. When geographic distances were standardized for both species, differing spatial patterns of population structure became apparent, with alewife exhibiting stronger population structure than blueback herring at all but the smallest of spatial scales (Figure 3).

Discussion

With some exceptions, we found significant differentiation among river spawning runs in both alewife and blueback herring, suggesting that the major river drainage is the appropriate level of management for these species. This result supports the findings of the ASMFC Stock Assessment, which found a general lack of correlation for temporal population trends among neighboring runs (ASMFC 2012).

Although genetic independence was the general pattern observed, in some cases we did not find significant genetic divergence among neighboring rivers. This result suggests that straying among neighboring watersheds has led to some degree of homogenizing gene flow (see also Palkovacs et al. 2008), but within larger-scale geographic boundaries. The location of these larger-scale geographic boundaries to gene flow (i.e., across which gene flow is extremely minimal) is what defines the higher-level population structure or stock structure. There is a high level of congruence between what pairwise genic tests identify as regions of gene flow and what Bayesian clustering methods identify as genetically distinguishable stocks. Thus, we have good confidence that we have identified the major genetic stocks for alewife and blueback herring in the USA portions of the species' ranges.

The designation of these stocks indicates that gene flow is not continuous across all parts of the species' ranges. There are geographic breaks in gene flow that should be recognized, as they represent the major

sources of intraspecific variation observed at neutral genetic loci. Importantly, these breaks in gene flow are not the same in alewife and blueback herring (Figures 1, 2) Both species show congruent Northern New England Stocks (ME, NH). However, the Southern New England Stock in alewife includes all of MA, RI, and CT to the Hudson River (NY), whereas the Southern New England Stock in blueback herring includes only MA and RI, with the Connecticut River (CT) belonging to the Mid-Atlantic Stock. In blueback herring, the Mid-Atlantic Stock extends from the Connecticut River (CT) to the Neuse River (NC). In alewife, the Mid-Atlantic Stock extends from the Delaware River (NJ) to the Chowan and Alligator Rivers (NC). In blueback herring, the Southern Stock extends from the Cape Fear River (NC) to the St Johns River (FL). This Southern Region is outside of the present range of the alewife, but it was not always so.

The alewife appears to be experiencing a range contraction at the southern end of its distribution, with populations extirpated from South Carolina, and now possibly extirpated from southern North Carolina. Based on annual NC WRC surveys of spawning adults conducted since 2006, alewives are possibly extirpated from the Cape Fear River, and at extremely low abundances in the Neuse and Tar-Pamlico Rivers (B. Wynne, *personal communication*) Larval sampling confirms these observations (A. Overton, *personal communication*). Marine bottom trawl surveys conducted by NMFS also indicate declines in alewife at the southern end of its distribution (ASMFC 2012).

Our results show greater IBD in alewife compared to blueback herring, suggesting that gene flow among runs is greater for the latter. This finding, together with our elucidation of stock structure boundaries for each species, has important implications for managing stocking practices. For example, stocking should not occur across major stock boundaries for either species. Higher straying rates for blueback herring make the effects of stocking across drainages perhaps less disruptive for natural population structure in this species; however, greater straying in blueback herring also makes natural re-colonization of watersheds more likely (and hence stocking perhaps less necessary to re-establish runs).

One of the utilities of genetic stock identification for anadromous species is the ability to assign marine-caught specimens back to natal spawning stocks. For species where all spawning runs are not significantly differentiated, it is difficult to consistently make reliable watershed-level assignments of natal origins. Thus, stocks can serve as biological units for the assignment of marine caught specimens such as those captured in fisheries (as bycatch, in the case of river herring).

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Table 1: Sample locations and sample sizes by year for alewife.

Code	Drainage	Latitude	2008	2009	2010	2011	2012	Total
EMAC	East Machias River, ME	44°41'27 N			58			58
STGEO	St. George River, ME	43°56'45 N			69			69
LAMP	Lamprey River, NH	43°03'59 N			47			47
MYST	Mystic River, MA	42°20'39 N			68			68
TRBO	Town Brook, MA	41°57'23 N				46		46
MON	Monument River, MA	41°43'24 N				49		49
GIL	Gilbert Stuart, RI	41°26'29 N				44		44
THAM	Thames River, CT	41°19'23 N		36				36
BRID	Bride Brook, CT	41°18'01 N		34				34
CON	Connecticut River, CT	41°16'58 N		7		26		33
QUIN	Quinnipiac River, CT	41°17'03 N		25				25
HOUS	Housatonic River, CT	41°10'10 N	13	25				38
MIAN	Mianus River, CT	41°00'58 N		25				25
HUD	Hudson River, NY	40°41'45 N		13			48	61
DEL	Delaware River, NJ	39°06'44 N				42		42
NAN	Nanticoke River, MD	38°10'08 N				58		58
RAP	Rappahannock River, VA	37°29'36 N				62		62
CHOW	Chown River, NC	37°12'33 N				54		54
ROA	Roanoke River, NC	35°55'22 N				49		49
ALL	Alligator River, NC					49		49

Table 2: Sample locations and sample sizes by year for blueback herring.

Code	Drainage	Latitude	2008	2009	2010	2011	2012	Total
EMAC	East Machias River, ME	44°41'27 N			57			57
STGEO	St. George River, ME	43°56'45 N			42			42
EX	Exeter River, NH	43°03'59 N			41			41
MYST	Mystic River, MA	42°20'39 N			66			66
MON	Town Brook, MA	41°57'23 N				50		50
GIL	Gilbert Stuart, RI	41°26'29 N				38		38
CON	Connecticut River, CT	41°16'58 N	34	62		46		142
HUD	Hudson River, NY	40°41'45 N		77				77
DEL	Delaware River, NJ	39°06'44 N				48		48
NAN	Nanticoke River, MD	38°10'08 N				24		24
RAP	Rappahannock River, VA	37°29'36 N				58		58
JAM	James River, VA	36°58'07 N				97		97
CHOW	Chown River, NC	37°12'33 N			12	58		70
ROA	Roanoke River, NC	35°55'22 N				50		50
NEU	Neuse River, NC	35°04'35 N				65		65
CF	Cape Fear River, NC	33°55'43 N				57		57
SAN	Santee River, SC	33°13'59 N				61		61
SAV	Savannah River, GA	32°02'53 N				51		51
ALT	Altamaha River, GA	31°18'53 N				52		52
STJ	St. John's River, FL	30°24'29 N				37		37

Table 3: Probability values for pairwise tests of genic heterogeneity among alewife populations from across the species' USA range. Instances of non-significant (p>0.05) genic heterogeneity are in bold.

	EMAC	STGEO	LAMP	MYST	MON	TBRO	GIL	THAM	BRID	CON	QUIN	HOUS	MIAN	HUD	DEL	NAN	RAP	CHOW	ROA
STGEO	0.0000	.																	
LAMP	0.0000	**0.5853**	.																
MYST	0.0000	0.0000	0.0000	.															
MON	0.0000	0.0000	0.0000	0.0000	.														
TBRO	0.0000	0.0000	0.0000	0.0000	0.0000	.													
GIL	0.0000	0.0000	0.0000	0.0000	0.0021	0.0000	.												
THAM	0.0000	0.0000	0.0000	0.0000	0.0004	0.0000	0.0259	.											
BRID	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0002	**0.2080**	.										
CON	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0239	0.0000	.									
QUIN	0.0000	0.0000	0.0000	0.0000	0.0002	0.0000	0.0033	**0.5115**	**0.0704**	0.0026	.								
HOUS	0.0000	0.0000	0.0000	0.0196	0.0000	0.0000	0.0001	**0.1761**	0.0022	0.0011	**0.0893**	.							
MIAN	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0001	.						
HUD	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0121	0.0000	0.0003	0.0135	**0.0618**	0.0000	.					
DEL	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0013	0.0000	0.0001	0.0036	0.0128	0.0000	0.0299	.				
NAN	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	**0.0765**	.			
RAP	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0056	0.0000	.		
CHOW	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	.	
ROA	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	**0.4563**	.
ALL	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	**0.2920**	**0.1349**

Table 4: Probability values for pairwise tests of genic heterogeneity among blueback herring populations from across the species' range. Instances of non-significant (p>0.05) genic heterogeneity are in bold.

	EMAC	STGEO	EX	MYST	MON	GIL	CON	HUD	DEL	NAN	JAM	RAP	CHOW	ROA	NEU	CF	SAN	ALY	SAV
STGEO	0.0005	.																	
EX	0.0000	0.0000	.																
MYST	0.0000	0.0000	0.0000	.															
MON	0.0000	0.0000	0.0000	0.0000	.														
GIL	0.0000	0.0000	0.0000	0.0000	0.0000	.													
CON	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	.												
HUD	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	.											
DEL	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0001	.										
NAN	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	**0.1264**	**0.6708**	.									
JAM	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0001	0.0000	**0.5709**	.								
RAP	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	**0.0720**	**0.0441**	**0.7936**	0.0009	.							
CHOW	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0001	**0.2726**	0.0004	0.0034	.						
ROA	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0007	**0.0910**	**0.4180**	**0.1168**	**0.0597**	0.0171	.					
NEU	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0247	0.0007	0.0132	0.0105	**0.6029**	.				
CF	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	.			
SAN	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	.		
ALY	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0056	.	
SAV	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0078	.
SJR	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000

Fig. 1: Proposed stock structure in alewife identified using the Bayesian clustering algorithm implemented in BAPS v.5.1 (Corander et al. 2006)**.**

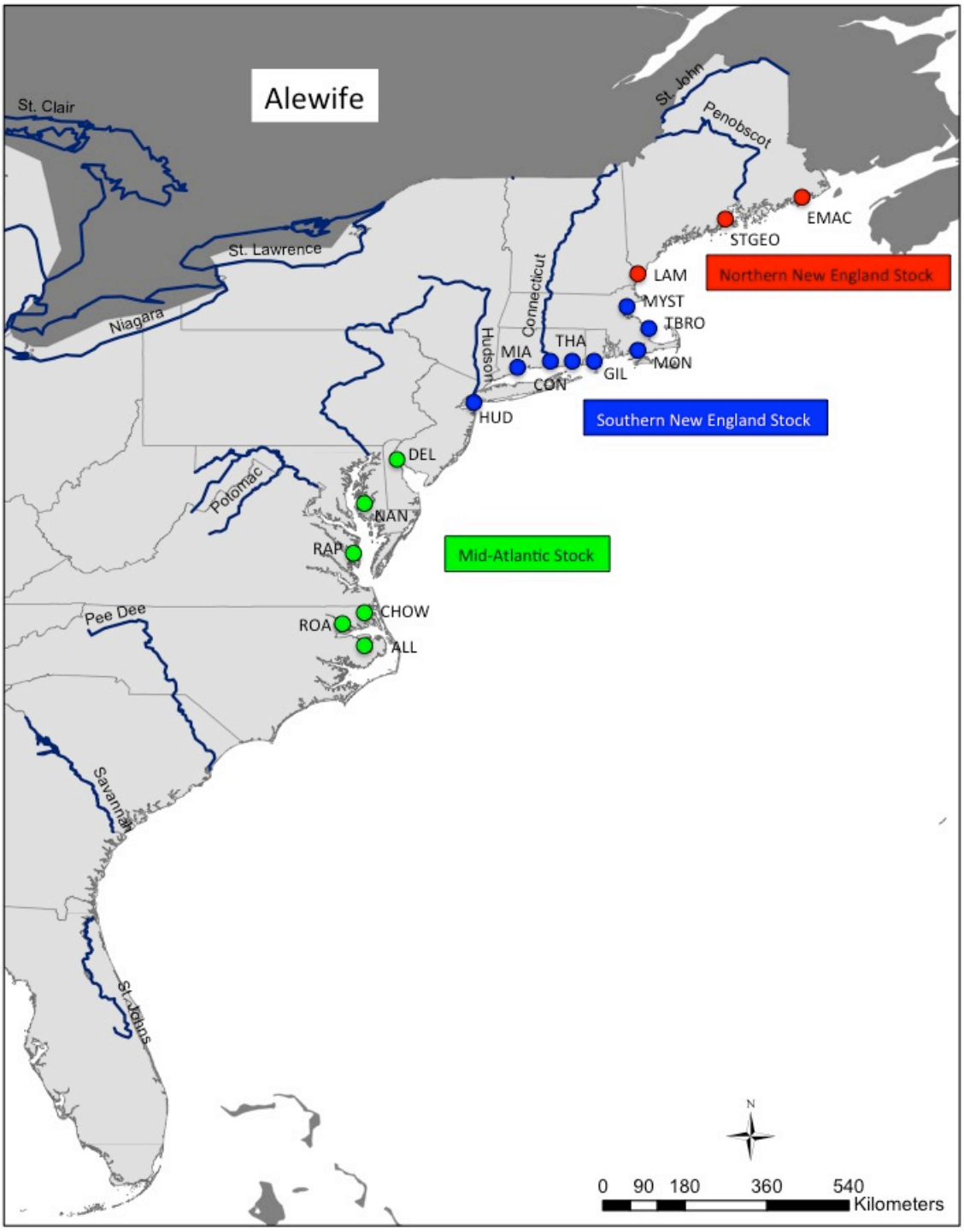

Fig. 2: Proposed stock structure in blueback herring identified using the Bayesian clustering algorithm implemented in BAPS v.5.1 (Corander et al. 2006).

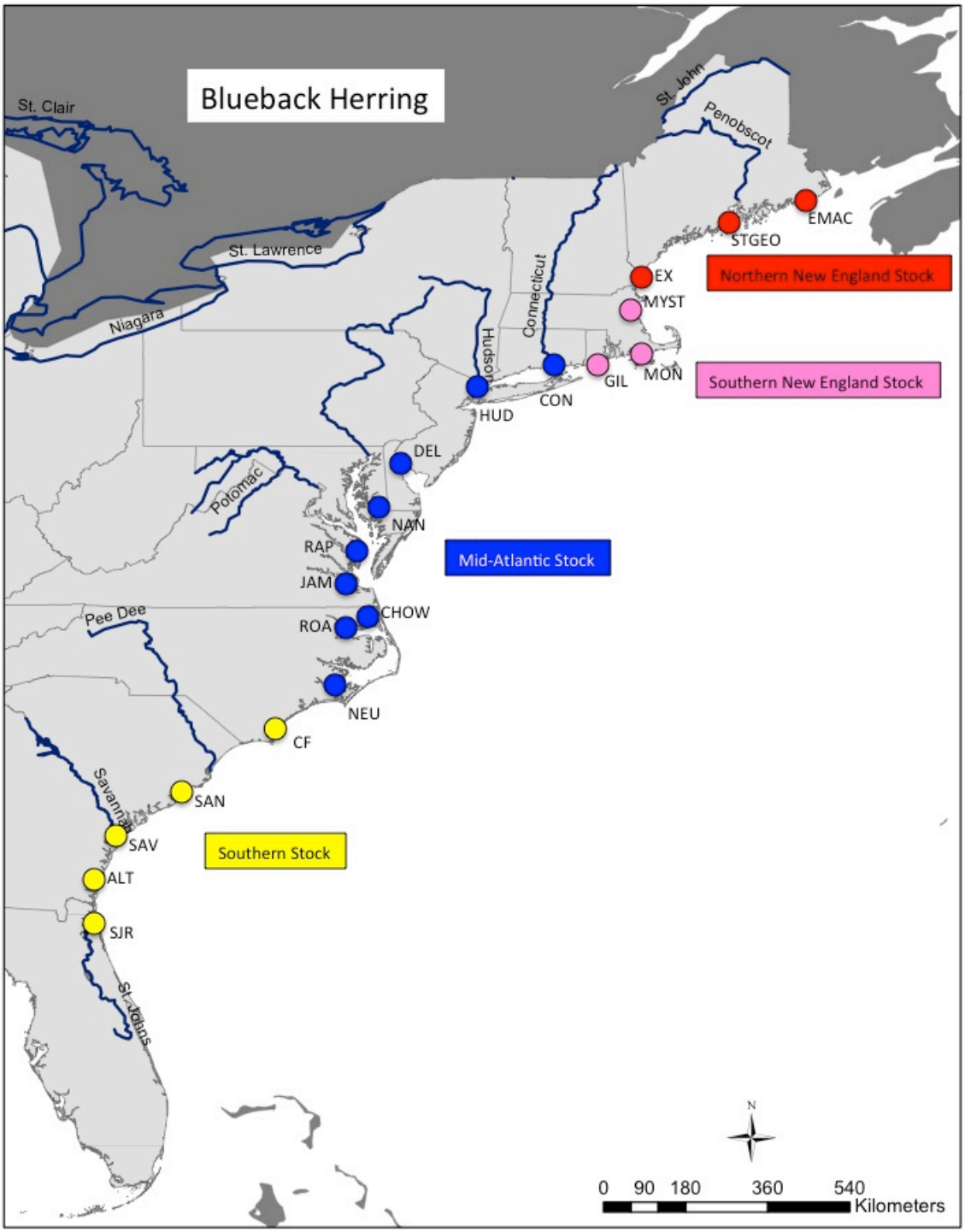

Fig. 3: Isolation by Distance (IBD) for alewife and blueback herring populations. IBD is stronger for alewife compared to blueback herring, suggesting greater gene flow via straying in blueback herring.